Plasmid DNA Purification Kit

(Catalog #K2802-200; 200 preparations; Store at 4˚C)

I. Introduction:

BioVision's plasmid DNA extraction and purification kit uses a unique system of magnetic beads and buffers to extract highly pure plasmid DNA from low or high copy plasmids. This kit does not involve the use of any toxic substances. It is safe, convenient, and suitable for high-throughput automated platforms. DNA purified by this kit can be used for a variety of downstream molecular biology applications such as cloning, PCR and more.

II. Kit Contents:

Component	K2802-200 200 preparations	Part Number
Buffer A	20 ml	K2802-200-1
Buffer B	20 ml	K2802-200-2
Buffer C	15 ml	K2802-200-3
Magnetic beads	1 ml	K2802-200-4
Wash solution	30 ml	K2802-200-5
Elution Buffer	20 ml	K2802-200-6

III. General Considerations and Reagent Preparation:

- Read the entire protocol before beginning the procedure.
- Incubate Buffer B in a 37˚C water bath before use until any white precipitate dissolves. Mix gently.
- Mix the magnetic beads to make a homogeneous suspension before use.
- Add 200 µl of RNase A (10 mg/ml) to Buffer A and 0.16 g NaOH to Buffer B before use.
- Add 80 ml absolute ethyl alcohol (user-provided) to wash solution and mix before use.
- If the volume of bacterial culture is increased, the volume of magnetic beads should be scaled-up proportionally.

IV. Bacterial DNA Extraction Protocol:

A. Lysis:

1. Take 1-2 ml (2-5 ml for Low copy plasmids) overnight bacterial culture in a tube and centrifuge at 12,000 x g for 2 min at 4˚C to collect cells. Remove supernatant completely. This can affect the purity of the plasmid DNA.
2. Add 100 µl Buffer A and completely resuspend cells making sure there are no clumps. Vortex if needed to loosen the pellet. Add 100 µl Buffer B and mix for 3 mins. until the solution becomes clear. Then add 75 µl Buffer C, mix by rotating and let it stand for 2 mins. to neutralize.
3. Centrifuge at 12,000 x g for 10 mins. and transfer the supernatant into a fresh tube.

B. Binding:

1. Add 5 µl re-suspended magnetic beads and 250 µl Isopropanol (user-provided) to the tube. Mix by inverting/rocking the tube for 5 mins. DO NOT Vortex.
2. Separate the beads by the magnetic separator (BioVision, Cat. 1999-1) and remove the clear supernatant (residual liquid).

C. Washing:

1. Add 500 µl wash solution, mix for 1-2 mins. Repeat magnetic separation and remove supernatant. Dry the beads at RT for 5 mins.

D. Elution:

1. Add 100 µl Elution buffer, pipette up and down slowly to mix, incubate at 65˚C for 10 mins. Rock the tube gently every 2-3 min.
2. Repeat magnetic separation and transfer the supernatant to a fresh tube carefully for downstream experiments.
3. Store the extracted DNA at –20˚C for future use. (Note: Generally, OD260 - OD320)/(OD280 - OD320) is between 1.7 ~ 2.0)

RELATED PRODUCTS:

EZLys(TM) Bacterial Protein Extraction Reagent (K8001-100,-500)
Ampicillin Sodium (2484-5G, -25G, -100G)
Carbenicillin disodium (2485-1G, -5G, -10G)
Ethidium Bromide (1203-10)
Plant Genomic DNA Extraction Kit (K2800-100)
Bacterial Genomic DNA Extraction Kit (K2801-100)
Viral DNA/RNA Extraction Kit (K2803-100)
Whole Blood Genomic DNA Extraction and Purification Kit (K2804-100)
Mitochondrial DNA Isolation Kit (K280-50)
Genomic DNA Isolation Kit (K281-50)

FOR RESEARCH USE ONLY! Not to be used on humans.

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